# American Journal of CLINICAL PATHOLOGY

## TECHNICAL SUPPLEMENT

Vol. 1	NOVEMBER, 1937	No.
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PUBLISHED BI-MONTHLY BY THE WILLIAMS & WILKINS COMPANY MOUNT ROYAL AND GUILFORD AVES., BALTIMORE, U. S. A.

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#### ANNOTATIONS, MINOR CONTRIBUTIONS, QUERIES

Under the above caption will be published from time to time comments, criticisms and suggestions on technical procedures; minor contributions such as laboratory aids and short cuts which are not considered sufficiently important to warrant a formal paper; and queries.

Obviously comments and criticisms should be signed; queries should be signed but names will be withheld on request; full credit will be given those who

contribute laboratory aids, short cuts and the like.

An attempt will be made to obtain answers from authoritative sources to the queries submitted. It must be emphasized that the views expressed in this department are not the opinions of any official body.

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# AN INEXPENSIVE, EASILY MADE, SAFETY RAZOR BLADE MICROTOME WITH TECHNIC ESPECIALLY USEFUL IN THE IMMEDIATE MICROSCOPIC DIAGNOSIS OF TISSUE\*

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Some years ago the discovery of what seems to be a new principle in microscopy¹ led to the development of a razor section technic in which thin staining on one side only of the section, in large measure took the place of thin sectioning. Several improvements in this new razor section technic were made and reported². Eventually the technic was so satisfactory that in examining over seven thousand malignant tissues at the Mayo Clinic, the diagnosis made upon sections cut with a razor, checked satisfactorily with the diagnoses made by four other pathologists using frozen sections in a little over 98 per cent of the cases studied³.

During the last four years the razor section method has been tested by me in the practical diagnosis of surgical tissues and has proved to be most helpful. In all cases the diagnoses made on razor sections have been controlled by frozen sections of the same tissue stained by Polychrome Methylene Blue or by hematoxylin and eosin or by both. In many biopsies the razor sections were better than the frozen sections. In a smaller number of cases I have preferred frozen sections. Although razor sections were sometimes unsatisfactory, they were never misleading.

Christeller4 reports that in one hundred and four cases in which

\*Read before the Sixteenth Annual Meeting of the American Society of Clinical Pathologists, Philadelphia, Pennsylvania, June 5, 1937.

The equipment necessary for this method (special cutting board, special slides, stain, etc.), may be obtained from Dr. Terry at a cost approximating \$3.00.

he used the older razor section technic, there were only three cases in which his diagnoses made on razor sections did not correspond with his diagnoses arrived at subsequently using other technic. But even in these three cases Christeller did not regard the razor section technic as misleading or at fault.

Hellwig<sup>5</sup> also has reported favorably on razor sections. In studying one thousand and thirty biopsies Hellwig found complete agreement between the diagnoses obtained by razor and by paraffin sections in 93.1 per cent. Malignancy and benignancy were correctly recognized by him in razor sections in 96.6 per cent of the cases. Hellwig prefers razor sections to frozen sections for the immediate diagnosis of biopsies.

It should be remembered that Christeller and Hellwig employed my older technic. If they could have used the technic about to be described, their results should have been even more satisfactory.

#### I. DISADVANTAGES

Perfection has never been claimed for the razor section method and I still control by frozen sections all diagnoses that I make on razor sections. Four disadvantages of the razor section method should be mentioned.

1. The method is not yet easily applicable to all tissues. If the tissues are extremely minute, extremely soft or friable, or opaque, or red, or black, it is usually better to use other methods.

2. Razor sections of unfixed tissue gradually decolorize the stain and the sections may have to be stained repeatedly if the pathological process is difficult to evaluate.

3. Even in sections of fixed tissue the stain in the superficial layer of razor sections gradually sinks into the depths and becomes unsatisfactory. It is, however, possible to restain sections several times before they become too saturated with stain to give satisfactory results.

4. The very ease with which razor sections can be prepared for microscopic examination may be regarded as a disadvantage, in that doctors who are not skilled in the interpretation of pathological processes may be tempted to give a diagnosis or express

an opinion on razor sections. Even technicians may try this. It should be remembered that the interpretation of razor sections requires as much experience, if not more experience, than the interpretation of good frozen sections or paraffin sections.

### II. ADVANTAGES OF RAZOR SECTION TECHNIC

Like the older technic, the newer razor section technic has the following advantages.

- 1. Wide applicability. The method is applicable to tissues recently fixed thoroughly in formalin. It is not equally applicable to tissues which have been fixed in formalin a long time. In many, but not in all, instances it is applicable to fresh unfixed tissue.
- 2. Dependability. Where the method is applicable, it is dependable. This statement is borne out by the experiences of Christeller<sup>4</sup> and Hellwig<sup>5</sup>.
- 3. High magnification. The highest powers of the microscope including the oil immersion lens, can be easily and satisfactorily used in studying razor sections. For oil immersion examination, mount the section on a glass slide stained side up, cover with a thin cover glass, and fill in the space below the cover with distilled water or tap water.
- 4. Few artefacts. Razor sections are so free from artefacts that this method can be used to check appearances thought to be artefacts in sections prepared by other methods.
- 5. Thorough. With no other method can tissue be so thoroughly examined in so short a time.
- 6. Emergencies. In experienced hands the method seems ideal for emergency examination.
- 7. Inexpensive. Razor sections are inexpensive. For the most part the razor section method uses things which are either already on hand in all well equipped laboratories or which can be procured at very little expense.
- 8. Noiseless. The method is practically noiseless. It uses no carbon dioxide and can be practiced in or near the operating room, in the doctor's office, or even in a private home.
  - 9. Quickly learned. The technical steps are so quickly learned

and are so easily carried out, that the pathologist becomes independent of a technician. He can cut and stain his own sections in less than sixty seconds.

10. Speed. The razor section method is the fastest known for preparing tissue for satisfactory examination under the microscope.

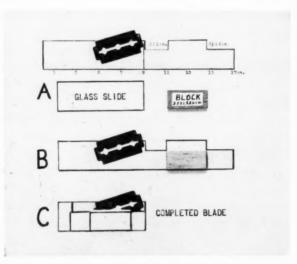


Fig. 1. A, Safety razor blade projects 1.1 cm. in slanting position over upper edge of sticky side of a double layer of adhesive plaster 17 cm. long. B, Razor is covered by glass slide. Block of wood added to adhesive between the two cut-outs. C, Adhesive to right of slide is fastened to block of wood, folded to left, and attached to glass slide. More detail in paper.

#### III. FOUR RECENT IMPROVEMENTS

1. Safety razor blade. Instead of using the relatively expensive old-fashioned biconcave razor requiring constant attention and labor to keep it in proper condition, inexpensive, double edged safety razor blades that fit into a Gillette razor are now employed in a holder that anyone can make quickly, easily, and inexpensively by following directions given in this paper (fig. 1).

2. Cutting board. A new cutting board of wood has been

devised which permits the cutting of sections of almost any desired thinness. Directions for making this cutting board are given later in this paper (fig. 2).

3. Immobilization. Instead of pinning blocks of tissue to a sheet of cork as was formerly done, the block of tissue is now immobilized by holding it beneath soft rubber. In addition the tissue is supported on three sides by wood (figs. 6 and 7).

4. Atomizer. The stain (Neutralized Polychrome Methylene Blue, Terry) is now applied to the surface of the section, not by

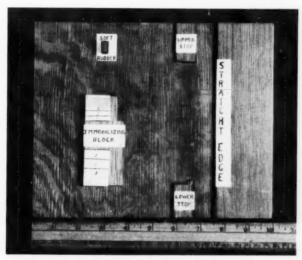


Fig. 2. Cutting Board Showing Straight Edge, Upper and Lower Stops, Immobilizing Block of Wood, and a Soft Rubber Block

a camel's hair brush as in the older technic, but by spraying the stain on the surface of the tissue, using a small, inexpensive atomizer. The section is mounted on a wet sponge so that any stain which goes over the edges, is at once diluted and in this way is prevented from staining the under surface of the section. Stain from the atomizer covers the surface of the section evenly and quickly and none of the stain is ever used twice or is contaminated by tissue juices or by tissue fragments picked up by the brush.

#### IV. ADVANTAGES OF THE IMPROVED RAZOR SECTION TECHNIC

1. Wider applicability. The newer and improved technic has even wider applicability than the older technic in that some of the tissues which formerly were difficult or impossible to section, can now be sliced more thinly and uniformly. With the older technic very friable tissues sometimes split apart when pins were thrust into them. Pins also failed to support properly soft, spongy tissues like lung and endometrial fragments. These tissues are better supported by the newer technic and the results are more satisfactory.

2. With the older technic the thinness of the sections depended in large part upon the skill of the one employing it. The newer technic is so mechanical that without unusual skill practically any desired thinness can be obtained.

To acquire the newer technic three things are necessary: (1) Secure the necessary equipment and supplies; (2) master the technical steps; (3) practice frequently what you learn.

#### V. EQUIPMENT AND SUPPLIES

The following equipment and supplies are desirable.

1. A good compound microscope with the usual lenses employed in pathological examinations.

2. Inexpensive, new double-edged safety razor blades. The ones I am using fit a Gillette razor and cost about one cent a piece.

3. Clean glass slides and cover slips. The cover slips should be of several sizes and some should be thin enough for use with the oil immersion lens.

4. Water proof or wet proof adhesive plaster 2.5 cm. wide. A roll five to ten yards long is convenient.

5. A cutting board of three ply hard wood panel or presdwood (fig. 2).

6. A rectangular piece of wood 9 x 4 x 0.6 cm. from the upper and longer right side of which a rectangular recess 2.5 x 0.5 cm. has been cut (fig. 2).

Beneath the left and longer side of this 9 x 4 x 0.6 cm. piece of wood are placed three thicknesses of adhesive plaster. It is convenient to use strips 15 cm., 14 cm., and 13 cm. in length by 2.5 cm. in width, applying first the 15 cm. strip flush with the left edge of the under surface of the block, then the 14 cm. strip, and finally the 13 cm. strip. The purpose of this adhesive plaster is to raise the right edge of the rectangular block from the cutting board so that the razor blade may slide freely beneath this block of wood.

7. Several pieces of soft rubber eraser about 1.5 x 0.7 x 0.7 cm. to be used in immobilizing tissue placed in the recess cut in the  $9 \times 4 \times 0.6$  cm. piece of wood.

8. A thin slice of fine grain sponge, known as "silk sponge." Slices 3 x 2.5 x 0.2 cm. are convenient. The slice of sponge is placed dry on the center of a glass slide and the ends of the sponge are fastened securely to the slide by adhesive plaster (fig. 11).

9. Two tumblers or glasses of clean cold tap water or distilled water.

10. A medicine dropper.

11. A pair of fine, sharp pointed forceps, capable of picking up a hair. The

spring in these forceps should be very delicate.

12. Stain. The new stain (Concentrated Neutralized Polychrome Methylene Blue, Terry) is not on the market. It is riper and more concentrated than the stain made according to my last published directions. For the present and until my latest directions are published, a free sample of the new concentrated stain can be obtained from me free of charge on request. If properly prepared, stain made by my older directions also gives satisfactory results provided it is used in the right concentration.

13. Special slides of wood which have in the center an oval hole measuring about  $3.5 \times 1.8$  cm. This hole is covered by a glass cover slip held in place at

either end by strips of adhesive plaster (fig. 16).

14. A 100-watt frosted Mazda electric light bulb provided with a shade.

15. An even stronger light is sometimes very helpful. I am using a lamp procured from E. Leitz of Wetzler. This is a special 6 volt, 5 amp. lamp provided with a variable resistance so that the intensity of the light can be varied in accordance with the thickness or opacity of the section. Hellwig<sup>5</sup> recommends a small arc light. A photoflood No. 1 bulb, may also be used.

16. A small inexpensive atomizer for spraying stain on the sections. My

atomizer cost about fifty cents.

17. A convenient recepticle for waste tissue.

Razor blade holder. The safety razor blade is double edged and is held in a slanting position to a glass slide by two thicknesses of adhesive plaster. In order to facilitate the use of this blade a block of wood is mounted on top of the glass slide and is held there by adhesive plaster (fig. 1).

To make this cutting instrument four things are needed:

- 1. Thirty-four centimeters of "wet proof" adhesive plaster 2.5 cm. wide.
- 2. A glass slide, about 7.6 x 2.5 x 0.1 cm., such as is used in all laboratories.

3. A sharp double edged razor blade.

4. A block of wood measuring 3.5 x 1.5 x 0.6 cm.

Cut the 34 cm. length of adhesive plaster in two strips, each 17 cm. long. To some clean, smooth, dry surface stick one of these strips and then add the other to the top of the first piece, aligning accurately all of the edges. Press the second piece of adhesive plaster firmly in contact with the first and then pull the double thickness of adhesive plaster away and place it in front of you with the sticky side up and with the long axis running from left to right. Mark on this sticky surface two points 9 cm. to the right of the left end of the strip. One of these points should be on the upper edge of the adhesive and the other

on the lower edge. With a pencil join the two points by a line which we will henceforth refer to as the "9 cm. line." To the right of this line two cut-outs are to be made from the upper edge of this adhesive plaster. The first cut-out is 1 cm. deep by 0.6 cm. broad and begins 1.5 cm. to the right of the 9 cm. line. The second cut-out is also 1 cm. deep but is 2.5 cm. long. It is cut from the extreme right end of the upper border of the adhesive strip. The two cut-outs are separated from each other by 3.5 cm.

With the adhesive plaster in front of you sticky side up and with its long axis running from left to right, place the razor blade on the adhesive as in figure 1. The right end of the blade should just touch the 9 cm. line, but the right end of the cutting edge of the blade should project 1.1 cm. over the upper edge of the adhesive plaster. When this is done the adhesive plaster bisects the right end of the hole in the center of the blade. The left end of the cutting edge of the blade should be flush with the upper edge of the adhesive plaster (fig. 1).

To hold the blade securely a glass slide is placed on top of it, the right end of the slide coinciding with the 9 cm. line. Align the upper and lower edges of the slide to coincide with the upper and lower edges of the adhesive plaster. When this is done, 1.4 cm. of adhesive plaster will project to the left of the left end of the slide. Bend this 1.4 cm. of plaster upward and to the right, fastening it neatly to the upper surface of the glass slide.

The wooden block is placed on the adhesive plaster between the two cutouts aligning the lower 3.5 cm. side of the block with the lower edge of the adhesive plaster. Press the block firmly in contact with the adhesive plaster. Keeping taut the plaster between the glass slide and the block, raise the right end of the block until it stands at right angles to the table and press the left end of the block, which is now the lower end, firmly in contact with the adhesive plaster. The block will now stand on this end and be perpendicular to the table. The upper end of the adhesive is bent sharply to the left over the upper end of the block and pressed firmly in contact. The adhesive plaster is now stuck to three sides of the block. Bend straight up the free end of the adhesive plaster and fold the adhesive which lies between the glass slide and the block upward and to the left over the right end of the glass slide. Pull the block and the adhesive to the left so that the side of the block not covered by adhesive plaster will lie on the glass slide above the razor. The adhesive plaster to the left of the block is then pressed firmly in contact with the glass slide. Finally, press downward the 3.5 cm. length of adhesive plaster between the two cut-outs and the block will be firmly fastened to the slide. Should any adhesive plaster project beyond the long sides of the slide, trim this off with a sharp knife and the blade is ready to use.

This blade is my standard, is used for all sections, and cuts a fairly thick slice of tissue, but a slice which as a rule can be properly transilluminated. With the cutting board sections of almost any desired thinness can be cut.

Cutting board. The cutting board is of convenient size, has a smooth upper

surface, a straight edge on the right side to guide the razor blade holder, and has stops above and below to limit the length of the stroke (fig. 2).

My board is made of four thin, smooth, rectangular blocks of presdwood of different sizes, but all have a thickness of  $0.6~\rm cm$ . Board number one is the cutting board proper and measures  $25~\rm x~20~\rm cm$ . This board is used with the longer side nearest the operator. Board number two is the straight edge, measures  $20~\rm x~6~\rm cm$ ., and after aligning the upper, the right, and the lower edges, is glued to the extreme right end of board number one. Boards three and four are the stops for the razor blade holder and are glued to board number one parallel to but  $2~\rm cm$ . to the left of the straight edge. The upper stop measures  $4~\rm x~2~\rm cm$ . The lower stop also measures  $4~\rm x~2~\rm cm$ . but from the left side of its upper end a shallow rectangular notch  $1.4~\rm cm$ . long by  $0.3~\rm cm$ . deep has been cut. Into this notch the immobilizing board is fitted when tissue is to be sliced. These stops are separated from each other by a distance of  $12~\rm cm$ . (fig. 2).

It is helpful to mark on the cutting board with water proof India Ink a rectangle which shows where the block of tissue should be mounted. The diagonal of this rectangle is easy to determine. Place the razor holder and the immobilizing blocks on the cutting board as in figure 6. Thrust a pin into the cutting board where the lower end of the cutting edge of the razor is flush with the slide. This marks the upper right corner of the diagonal. A pin thrust into the cutting board in the corner of the recess in the immobilizing board gives the lower left corner of the rectangle. Complete the rectangle. It is shown in figure 2 and should measure about 1.3 x 0.7 cm.

To cut thinner sections cover the rectangular space with a single layer of adhesive plaster and mount the block of tissue on this adhesive plaster. Still thinner sections can be cut by adding additional thicknesses of wax paper beneath the block of tissue to be cut (fig. 5).

#### VI. TECHNICAL STEPS

These are almost the same as for the older technic described by me in 19304.

1. Selection. By inspection and palpation select that part of the tissue which is most apt to contain the pathological changes that should be studied microscopically.

2. Excision. The area selected is excised and for best results should not exceed 1 x 0.5 x 0.5 cm.

3. Wetting tissue. Before sectioning the tissue it is advantageous to wet it with a few drops of water in which a little sodium bicarbonate has been dissolved. Experience shows that tissue can be sectioned easier if it is wet. The sodium bicarbonate is used to prevent the razor from rusting. It is convenient to have a small amount of the bicarbonate in an open vessel and to dip the block of tissue in this solution before placing it on the cutting board (fig. 3).

4. Immobilization. After the razor in its holder is placed on the cutting board with the right side of the slide in contact with the left side of the straight edge



Fig. 3. Wetting the Block of Tissue with Sodium Bicarbonate and Draining

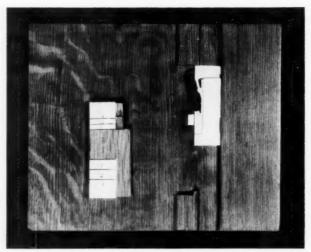


Fig. 4. Arrangement for Cutting Relatively Thick Sections.

Tissue on Cutting Board against Razor

and with the upper end of the razor against the upper stop, the block of moist tissue (fig. 3) is placed against the slide just below the lower end of the razor

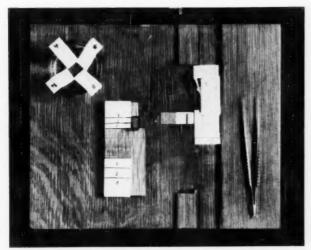


Fig. 5. Arrangement for Cutting Thin Sections

Tissue mounted on top of wax paper that covers one layer of adhesive plaster stuck to cutting board. Other thicknesses of wax paper shown in dish on cutting board.

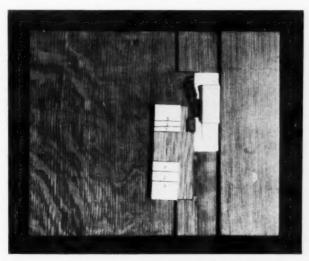


Fig. 6. Tissue Supported by Immobilizing Block and Covered by Soft Rubber Block. Razor Holder against Upper Stop and against Straight Edge

edge (fig. 4). Next the lower end of the rectangular immobilizing block is fitted into the notch of the lower stop, and the recess in its upper right corner is fitted against the block of tissue (fig. 6). The immobilizing block is so held along the left edge by two or three fingers of the left hand that it will not interfere with the subsequent movement of the razor. The immobilization is completed by placing the block of soft rubber on top of the tissue and holding this block gently against the tissue with the middle finger of the left hand (fig. 7). It is important not to exert too much pressure on this rubber block.

5. Slicing. The razor blade holder has already been placed in contact with the upper stop and with the left edge of the straight edge. With the thumb and second finger of the right hand grasp the block attached to the razor blade holder. The first finger of the right hand exerts pressure downward on the block, keeping the holder in close contact with the cutting board (fig. 8). The razor blade can be moved downward easily, keeping the right edge of the holder in contact with the left edge of the straight edge. This simple movement slices the tissue (fig. 9).

After slicing the tissue place the soft rubber on top of the rectangular immobilizing block and move both to the left. Then pick up the razor blade on which the block of tissue rests and place it to the right on the straight edge (fig. 10).

6. Transferring section to sponge. When the razor is raised the section frequently clings to the under surface. If it does so, wipe the section gently on the thin slab of wet sponge attached to a glass slide by adhesive tape. This sponge must be thoroughly wet with water before the tissue is placed on it. Should the tissue not adhere to the razor, pick it up gently by the edge with forceps and transfer it to the sponge taking care not to turn the section over (fig. 11). The side uppermost on the cutting board will have a smooth surface as it was next to the razor. It is this smooth surface that should be washed and stained.

7. First washing. After the tissue is transferred to the sponge, the upper surface is gently washed with water which is spurted on the section from the medicine dropper (fig. 12). Before staining be sure that the sponge is as full of water as possible.

8. Staining. To stain razor sections some of the Concentrated Neutralized Polychrome Methylene Blue, Terry, is diluted with nine times its volume of distilled water. This makes a one in ten dilution and is used in a small atomizer. For razor sections dilute stain is to be preferred to the concentrated as there is less likelihood of overstaining the sections or of accidently staining them on both sides. In applying the stain the nozzle of the atomizer is held close to the surface of the tissue, but does not touch it (fig. 13). The stain is sprayed gently on the surface and any excess that runs over the side is immediately diluted by the water in the sponge. In this way the tissue is stained on one side only. This is highly important as the section must subsequently be transilluminated.

Most sections are stained satisfactorily in one to five seconds. If the tissue fails to be stained sufficiently at the first attempt, spray the surface a second

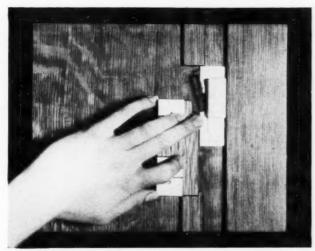


FIG. 7. TISSUE AND IMMOBILIZING BLOCK HELD DOWN BY LEFT HAND

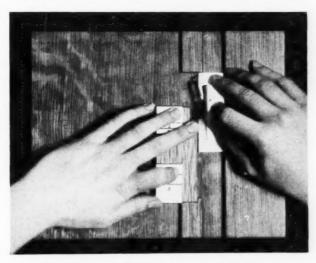


FIG. 8. RAZOR GRASPED WITH RIGHT HAND

time or even a third time. Should the one to ten dilution act too rapidly, dilute the stain still further. A dilution of one to twenty usually stains sec-

tions satisfactorily in five seconds or less. With the highly concentrated stain I am now making, the one to twenty dilution is the one I use on razor sections.



Fig. 9. Slicing the Tissue by Moving the Razor Downward

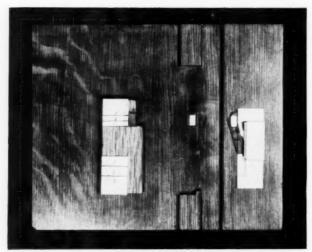


Fig. 10. Slice of Tissue in Rectangle. Immobilizing Blocks, Moved to the Left. Razor with Block of Tissue on Upper Surface, Moved to the Right

9. Second washing. As soon as the tissue is satisfactorily stained, wash the stain from the upper surface by gently spurting water on the tissue, using the medicine dropper (fig. 14).

10. Mounting. The tissue is conveniently mounted on the under surface of the cover glass attached to one of the thin wooden special slides (figs. 15 and

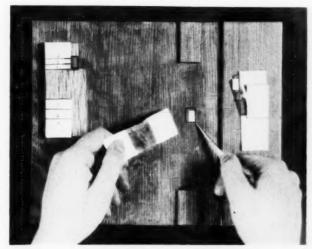


Fig. 11. Picking up the Tissue to Transfer It to a Wet Piece of Sponge

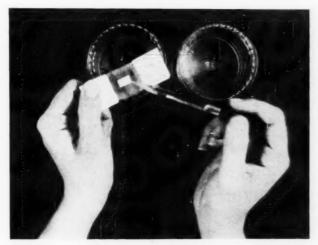


Fig. 12. First Washing of Slice of Tissue on Wet Sponge. Saturating Sponge with Water

16). These slides have an oval hole through the center. To this slide a cover glass has been attached at either end by a strip of adhesive plaster. Before

mounting, this slide is turned over so that the cover glass is below. The stained and washed section is now placed stained side down in contact with the cover



Fig. 13. Atomizer Spraying Dilute Stain on Tissue Supported by a Wet Sponge

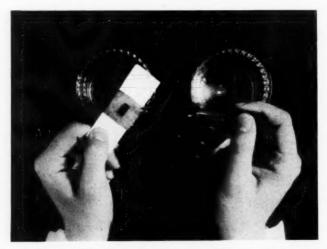


Fig. 14. Second Washing of Tissue to Remove Excess Stain

glass (fig. 15). On turning the slide over so that the cover glass is up, the specimen will stick to the cover with the stained side up and is ready for microscopic examination (fig. 16).

Razor sections cut by the improved technic are so thin and so nearly planeparallel that the special slides are no longer as necessary as formerly.

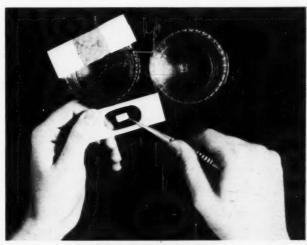


Fig. 15. Mounting the Tissue Stained Side down on Cover Glass Attached to a Special Wooden Slide Turned Upside Down

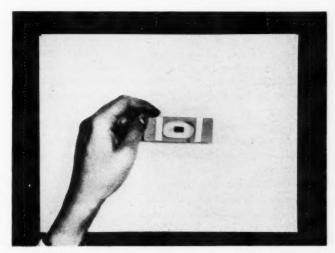


Fig. 16. Slide Turned Back to Normal Position Showing Section Stained Side up Clinging to Under Surface of Cover Glass. Section Ready to be Examined

Good thin razor sections stained on one side only may be mounted stained side up on ordinary glass slides and covered with a thin cover glass. After the

space beneath the cover is filled with distilled water or with tap water, these sections are ready for examination. Nevertheless, I usually prefer to mount my razor sections beneath covers attached to the special wooden slides already described.

11. Transillumination. To transilluminate razor sections light is required which is stronger than that used for ordinary thin, cleared sections of tissue. A 100-watt frosted Mazda bulb will suffice for most tissues. But occasionally the density of the section is so great, or the color is so dark, or red, that a stronger light is necessary.

#### VII. PRACTICAL POINTS

1. In modern razor section technic thin staining largely takes the place of thin sectioning. But the thin staining must be on one side only of the tissue and the side stained must be as smooth as possible. The stained side is always mounted in contact with the cover glass.

2. To cut a smooth surface, the razor blade must be smooth and superlatively sharp. Do *not* use discarded razor blades. They are usually lacking in sharpness and frequently are roughened due to rust.

3. The sectioning of fixed tissue is better if both the razor and the tissues are wet. After each use dry the blade at once to avoid rust.

4. Whenever the sectioning is disappointing, suspect that the razor blade is not sharp enough and try a new blade or turn the blade around and use the other edge provided it is sharp and not rusted.

5. Thin sectioning. If sections thinner than the standard thickness of two layers of adhesive plaster are desired, these thinner sections may be obtained by mounting the tissue block on one thickness of adhesive plaster before sectioning. Very thin sections have been obtained by mounting the block to be cut on one or more layers of wax paper placed on top of a single layer of adhesive plaster (fig. 5).

Although in thinness razor sections are not to be compared favorably with frozen sections, occasionally razor sections of fixed tissue have been cut which were thin enough to stain with hematoxylin and eosin, clear, and preserve as permanent specimens.

6. If the tissue is friable or extremely soft and if difficulty is encountered in picking up the sections after they are cut, it is convenient to mount the tissue on a single layer of wax paper before sectioning. The section will cling to the razor blade or to the paper. If it clings to the razor blade, the section can be wiped off gently on the wet sponge and stained. After staining the section can be mounted from the sponge or it can be floated free in the water and mounted from water on a slide and covered with a cover and examined.

If the section clings to the wax paper, it is easier to handle. Wet the section and paper well and then without removing the section from the paper, stain only the upper surface of the section. Avoid using an excess of stain.

The section can be washed gently and the paper trimmed so that it can be mounted with the section on the under surface of the cover on one of the special wooden slides.

7. The more opaque the tissue, the thinner the sections must be cut and the stronger they must be transilluminated.

8. Very red or very black tissues are especially difficult. The red ones because red absorbs the blue in the light; the black sections because black absorbs all of the light. Red and black tissues must be cut especially thin and must be strongly transilluminated.

9. Tissues stain best when perfectly fresh or when recently thoroughly fixed in formalin. Tissues fixed for a long time in formalin are much less satisfactory.

10. Partial fixation. If fresh tissues are so soft, so mucinous, or so porous that good results are seemingly difficult or impossible to obtain, it is sometimes advantageous to fix them partially by immersing small blocks of these tissues in boiling water for about eight to ten seconds. This heating makes sectioning much easier, but if it is overdone the staining is unsatisfactory.

#### CONCLUSION

The advantages of razor sections are so numerous and so real that it seems that every pathologist should give the technic a thorough trial and should use it, not in place of other methods, but in connection with them. In this way only will the true value of razor sections be appreciated.

#### REFERENCES

- A New Principle in the Microscopic Examination of Fresh Unfixed Tissue. Proceedings Weekly Staff Meeting Mayo Clinic, 1: No. 37, 209. December, 1926.
- (2) Terry, Benjamin T.: Improvement in Technic and Results Made in Examining Microscopically by the Razor Section Method 2000 Malignant Tissues. Jour. Lab. and Clin. Med., 14: No. 6, 519. March, 1929.
- (3) Terry, Benjamin T.: Rapid Diagnosis of Malignant Tumors. Texas State Jour. Med., 26: 434-439. October, 1939.
- (4) Christeller, Erwin: Erfahrungen mit der Verbesserten Histologisch-Diagnostischen Schnell-Methode nach Terry. Klin. Wehnschr., 7: 448–450. 1928.
- (5) Hellwig, C. Alexander: Tissue Diagnosing During Operation: Reliability of Terry's Supravital Technic in 1,030 Biopsies. Surg., Gynecol. and Obstet., 61: No. 4, beginning at page 494. October, 1935.

## PUBLISHED PROCEDURES RECOMMENDED FOR TRIAL

# A DEVICE TO FACILITATE THE IMPREGNATION OF RETICULIN FIBRILS IN PARAFFIN SECTIONS

A. H. T. Robb-Smith, J. Path. & Bact., 45: 312. 1937

In order to prevent the frequent occurrence of detachment of sections from the slides in the silver impregnation of sections the following method was devised. Essentially it consists of staining paraffin sections without fixing them to slides, just as celloidin sections are stained.

Sections are cut in the usual way and floated out in warm water. They are transferred from one solution to another using a slide instead of a section lifter. Manipulation with a camel hair brush prevents the breaking or tearing likely to occur when a glass or metal needle is used. Sections are *floated* on all solutions.

After the final step in the silver impregnation the sections are attached to slides with or without albumin, thoroughly dried, the paraffin removed, counterstained if desired, dehydrated and mounted.

A slightly modified Foot-Menard method proved constant in results with material fixed in various ways.

#### Procedure:

- 1. 10 per cent ammonium hydroxide: 15 minutes.
- 2. Wash in three changes of distilled water.
- 3. 0.25 per cent potassium permanganate: 5 minutes.
- 4. Wash in distilled water for a few minutes.
- 5. 1.5 per cent oxalic acid until brown color disappears.
- 6. Wash in four changes of distilled water.
- 7. 5 per cent silver nitrate: 1 hour at room temperature.
- 8. Wash in two changes of distilled water.
- 9. Ammoniacal silver: 15 minutes at room temperature.
- 10. Wash in three changes of distilled water.
- 11. 30 per cent formalin: 3 minutes.
- 12. Wash in tap water.
- 13. Tone if desired in 0.2 per cent gold chloride: 3 minutes.
- 14. Fix in 5 per cent sodium thiosulphate: 1 minute.
- 15. Wash and mount.

Ammoniacal silver: To 8 cc. of 10 per cent silver nitrate 6 drops of 10 per cent sodium hydroxide are added. Strong ammonia is added drop by drop until the precipitate is just dissolved. Make up to 28 cc. with distilled water. The resulting solution should not smell of ammonia.

# AN APPLICATION OF BLOOD STAINING TO FORMALIN FIXED TISSUES

Helen Campbell Wilder, J. Tech. Methods & Bull. Int. Assoc. Med. Mus., 14: 68. 1935

1. Xylol.

2. Absolute methyl alcohol 2 changes.

3. Cover slide with a freshly filtered saturated solution of Jenner's stain (National Aniline & Chemical Co., Inc., New York, N. Y.) in methyl alcohol: 3 minutes.

4. Add an equal amount of distilled water: 1 minute.

5. Plunge, without washing, into a Coplin jar of dilute Giemsa solution (1 drop of Giemsa's Spirochete Stain from Hynson, Westcott and Dunning, Baltimore, Md., to 1 cc. of distilled water): 45 minutes.

 Rinse and differentiate in acidulated distilled water (1 drop of glacial acetic acid to 5 cc. of distilled water).

7. Rinse in pure distilled water.

- 8. Dehydrate quickly in 95 per cent alcohol and two changes of absolute alcohol.
  - 9. Clear in xylol and mount in Canada Balsam.

-Recommended by W. S. Thomas.

# RAPID METHOD FOR PREPARING AND STAINING BONE MARROW

E. M. Schleicher and E. A. Sharp, J. Lab. & Clin. Med., 22: 949. 1937

By means of a syringe and needle (the authors have devised their own) 0.5 cc. of marrow is withdrawn and discharged into a paraffin lined tube containing heparin and centrifuged. The excess of plasma is removed and remaining plasma and cells mixed with a pipette. Films are made as with blood and stained as follows:

- Place slide in 0.3 per cent May-Grünwald stain in methyl alcohol for 3 minutes.
- 2. Without washing place slides in a dilute May-Grünwald stain (0.3 per cent alcoholic May-Grünwald stain and buffer solution pH 6.8 equal parts): 1 minute. This mixture should be freshly made.
- Cover slide with 3 cc. of dilute Giemsa's stain (one drop of stock Giemsa's Stain to 1 cc. of buffer solution): 5 minutes.
- Rinse with neutral distilled water, tilting the slide until water comes away clear. Place in vertical position to dry.

# A MODIFICATION OF THE SILVER IMPREGNATION METHOD FOR THE STAINING OF LATTICED FIBERS

VICTOR A. ZHOOKLIN, J. Lab. & Clin. Med, 22: 1284. 1937

1. Fix in formaldehyde.

2. Wash in running water one to two minutes. If fixation has been prolonged wash from one to two hours.

3. Frozen sections or decerated paraffin sections are transferred from distilled water to 1:100,000 to 1:50,000 potassium permanganate for two to thirty seconds depending on character of tissue and duration of fixation.

4. Ammoniacal silver: 2 to 3 minutes. (Ammoniacal silver: To 20 per cent silver nitrate solution add strong ammonia drop by drop until the precipitate is just dissolved, then add two or three drops in excess so that the solution has a perceptible odor of ammonia.)

5. Pass through distilled water to a freshly prepared 1 to 1.2 per cent solution of formalin pH 7. Keep sections in motion.

The degree of impregnation is controlled under the microscope.

6. Wash in water, dehydrate, and mount in balsam. If desired, sections may be toned in gold chloride as usual and counterstained.

## ANNOTATIONS, MINOR CONTRIBUTIONS, QUERIES

Modification of Gram Method. By adding sufficient Tr. Iodine to the absolute alcohol to make a light brown color washing out of the stain from Gram positive organisms is prevented. J. A. de Louriero, J. Lab. & Clin. Med., 22: 638, 1937.

Comparative Study of Dehydration. The paraffin method has frequently been criticised because of its hardening and shrinking effect on tissue. The author believes this distortion is due to the dehydration and not to the immersion in melted paraffin. An experimentally controlled series of various tissues was dehydrated in different dehydrating agents, dioxan, iso-butyl alcohol, and ethyl alcohol with chloroform. Except for the dehydration the tissues were treated identically. In every case, the dioxan proved to be a better dehydrating reagent with less shrinkage and brittleness than any of the others. Ethyl alcohol with chloroform produced the greatest degree of distortion. (Author's abstract.). Thelma T. Baird, Stain Tech., 11: 13, 1936.

BLOOD CULTURES USING PATIENTS' BLOOD AS SOLE MEDIUM. The authors defibrinate 10 cc. of the patients' blood in a 50 cc. flask containing glass beads and incubate without adding culture media of any kind. Their results were better than when the ordinary methods were used. Anna D. Delaney and Frances Guthrie, J. Lab. & Clin. Med., 22: 721, 1937.

ELECTRIFICATION OF PARAFFIN RIBBONS. A vessel of boiling water in the sectioning room will furnish a sufficient degree of humidity to allow the electric charge in sections to be dissipated and eliminate altogether the difficulty of handling. C. Wilson and J. S. Hockaday, Science, 82: 306, 1935.

Index photographed at the beginning for the convenience of the microfilm user.